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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
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EXAMINER	
DOWELL, PAUL THOMAS	

ART UNIT	PAPER NUMBER
1632	

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Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/743,391	HIRST, TIMOTHY RAYMOND	
	Examiner	Art Unit	
	Paul Dowell	1632	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *09 February 2006*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-4,6-8,10-18,21,23 and 24* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-4,6-8,10-18,21,23 and 24* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☒ The specification is objected to by the Examiner.

10)☒ The drawing(s) filed on *11/1/04* is/are: a)☒ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☒ None of:

1.☒ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date *12/22/2003*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: _____.

DETAILED ACTION

Election/Restrictions

The restriction requirement of 12/8/2005 is withdrawn in part. Specifically, Examiner has withdrawn the requirement for Applicant to elect either a mutant form or EtxB or a mutant form of CtxB as put forth on page 3 of the restriction requirement.

In the response of 2/9/2006 Applicant has elected claims 1-18, 21 and 23-25 (group I) and has elected the agent identified as peptide or protein of interest. Claims 1, 2, 10-15, 17 and 21 have been amended. Claims 5, 9, 19, 20, 22 and 25 have been canceled. Because Applicant did not distinctly and specifically point out the supposed errors in the restriction requirement, the election has been treated as an election without traverse (MPEP § 818.03(a)). The restriction requirement of 12/8/2005, modified as put forth herein above, is made FINAL.

Claims 1-18, 21 and 23-25 are pending and under examination in the instant office action.

Priority

Acknowledgment is made of applicant's claim for foreign priority based on an application filed in The United Kingdom on 6/22/2001. It is noted, however, that applicant has not filed a certified copy of the 01153823.4 application as required by 35 U.S.C. 119(b).

Specification

The specification is objected to because page 46, Table I lists peptides longer than 10 amino acids that are not properly identified by a SEQ ID NO.

Claim Objections

Claims 1, 12, 14, 15, 17 and 21 are objected to because of the following informalities: the terms EtxB and CtxB are not defined at the first recitation of said terms. Appropriate correction is required.

Claim Rejections - 35 USC § 101

35 U.S.C. 101 reads as follows:

> Whoever invents or discovers any new and useful process, machine, manufacture, or composition of matter, or any new and useful improvement thereof, may obtain a patent therefor, subject to the conditions and requirements of this title.

Claims 1, 14, 15 and 17 are rejected under 35 U.S.C. 101 because the claimed recitation of a use, without setting forth any steps involved in the process, results in an improper definition of a process, i.e., results in a claim which is not a proper process claim under 35 U.S.C. 101. See for example *Ex parte Dunki*, 153 USPQ 678 (Bd.App. 1967) and *Clinical Products, Ltd.* v. *Brenner*, 255 F. Supp. 131, 149 USPQ 475 (D.D.C. 1966). Claims 2-4, 6-8, 10 and 11 depend from claim 1; claim 16 depends from claim 15; and claim 18 depends from claim 17. As such, claims 2-4, 6-8, 10, 11, 16 and 18 are likewise rejected.

Claim Rejections - 35 USC § 112

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1-4, 6-8, 10-18, 21, 23 and 24 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for:

An *in vitro* method of delivering a peptide to a cell expressing GM-1 ganglioside receptors on the surface of said cell comprising contacting said cell with a mutant B-subunit of *E. coli* heat labile enterotoxin (EtxB) or a mutant B-subunit of *Vibrio cholerae* cholera toxin (CtxB) covalently linked to said peptide, said mutant EtxB or mutant CtxB comprising one of the following point mutations within the region spanning amino acid residues E51 to I58 of the β4-α2 loop of EtxB or CtxB: CtxB(E51A), CtxB(Q56A), CtxB(H57A) and EtxB(H57S), thereby delivering said peptide into said cell; and a kit comprising said mutant EtxB or mutant CtxB covalently linked to said peptide.

does not reasonably provide enablement for:

A method of delivering <u>any</u> agent to <u>any</u> target cell comprising contacting said cell with <u>any</u> mutant EtxB or <u>any</u> mutant CtxB, thereby delivering said agent to said cell resulting in treatment of <u>any and all</u> disorders and diseases.

The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims.

While determining whether a specification is enabling, one considers whether the claimed invention provides sufficient guidance to make and use the claimed invention, if not, whether an artisan would have required undue experimentation to make and use the claimed invention and whether working examples have been provided. When determining whether a specification meets the enablement requirements, some of the factors that need to be analyzed are: the breadth of the claims, the nature of the invention, the state of the prior art, the level of one of ordinary skill, the level of predictability in the art, the amount of direction provided by the inventor, the existence of working examples, and whether the quantity of any necessary experimentation to make or use the invention based on the content of the disclosure is "undue" (In re Wands, 858 F.2d 731, 737, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988)).

Furthermore, the USPTO does not have laboratory facilities to test if an invention will function as claimed when working examples are not disclosed in the specification, therefore, enablement issues are raised and discussed based on the state of knowledge pertinent to an art at the time of the invention, therefore skepticism raised in the enablement rejections are those raised in the art by artisans of expertise.

The instant claims are very broad in scope and encompass a method of treatment comprising delivering an agent to a target cell, wherein said agent is delivered by contacting said cell with a mutant EtxB or mutant CtxB, wherein said mutant EtxB or mutant CtxB has a mutation in the region spanning amino acid residues E51 to I58 of the β4-α2 loop of EtxB or CtxB.

The specification contemplates that the compositions of the present invention can be administered by direct injection, parenteral, muscoal, intramuscular, intravenous, subcutaneous, intraocular or transdermal administration (page 25, lines 4-12). The specification discloses laundry lists of delivery vehicles (page 25, lines 7-12) and of disorders and/or diseases to be treated by the claim methods (page 26, line 22 to page 27, line 10). For example, the specification recites a long list of infectious diseases to be treated including HIV-1 and HIV-2 (page 26, line 26) and a long list of cancers to be treated including melanoma and breast cancer (page 27, lines 7 and 9). The specification discloses results of experiments where intranasal or subcutaneous immunization of mice with wild type CtxB resulted in an immunological response to CtxB while immunization with mutant CtxB(H57A) led to a marked reduction in the immunological response (Example 4e, page 43). The specification discloses that EtxB(H57A) covalently conjugated to a 19mer peptide stimulated class I-restricted antigen presentation when incubated with cultured JAWS II dendritic cells as evidenced by IL-2 production from a specific T-cell hybridoma as a read out (page 55, line 30 to page 56, line 12).

However, the specification provides no working examples or specific guidance as to how an artisan would make and use the claimed invention to treat any one specific disease or disorder. The specification discloses results from one experiment where EtxB(H57A) covalently conjugated to a 19mer peptide stimulated class I-restricted antigen presentation when incubated with cultured JAWS II cells (page 56). Such a disclosure is not deemed sufficient to enable the scope of the instantly claimed

invention. In particular, the specification fails to establish a nexus between the claimed methods and treatment of any specific disease or disorder.

The art of record at the time of the invention teaches that peptide/protein agents conjugated to non-mutant CtxB can be administered to rodents to ameliorate experimentally induced autoimmune disorders. For example, Bergerot et al **(Proceedings of the National Academy of Sciences, 94:4610-4614, 1997)** teaches that human insulin conjugated to CtxB, when administered orally to diabetic NOD mice, delayed the onset of diabetes and reduced insulinitis in said mice (page 4611, col. 2, paragr. 4 to page 4612, col. 2, line 11). Sun et al **(Proceedings of the National Academy of Sciences, 93:7196-7201, 1996)** teaches that myelin basic protein conjugated to CtxB, when administered orally to Lewis rats before subcutaneous challenge with myelin basic protein, protects said rats from developing myelin basic protein-induced experimental autoimmune encephalomyelitis (page 7191, col. 2, paragr. 4 to page 7198, col. 1, line 13). However, both Bergerot and Sun teach methods using non-mutant CtxB that exhibits immunomodulatory activity (i.e. the non-mutant CtxB alone, irrespective of any conjugated or co-admistered antigen), unlike the mutant EtxB/CtxB taught by the instant specification.

Plant et al **(Current Topics in Medicinal Chemistry, 4:509-519, 2004)** teaches the state of the art and reviews the teachings of Bergerot and Sun (page 512: col. 1, paragr. 3 to col. 2, paragr. 2). Plant recognizes first, that antigens must be conjugated to toxin B subunits in order to elicit a biological response (page 512, col. 2, paragr. 2, lines 4) and second, that, "The finding that mucosally or parenterally administered EtxB or

parenterally delivered CtxB alone can prevent or treat autoimmune disease indicates that the B subunits have immunomodulatory activity beyond their ability to act as delivery vehicles" (page 513, col. 1, paragr. 1, lines 105). Thus, the teachings of Bergerot and Sun, and said teachings as summarized by Plant, indicate that toxin B subunits must possess immunomodulatory activity and must be conjugated to antigens in order to mediate a biological response in animal models of autoimmune disorders. As such, an artisan would experience undue experimentation to practice the claimed invention, for example, to elicit a biological response in animal models of autoimmune disorders, because it would be unpredictable whether mutant EtxB/CtxB lacking immunomodulatory activity would be operant and it would be unpredictable whether antigens or agents not conjugated to EtxB/CtxB would be operant.

Further, the breadth of the instant claims is such that they read on methods of treating any and all diseases and disorders. Claim 16 further limits the disease or condition of claim 15 to a viral infection or a cancer. Claim 16 reads on any viral infection or any cancer. The claims are not enabled as such because the specification fails to establish a nexus between the claimed methods and treatment of any specific disease or disorder, let alone any viral infection or any cancer. The specification does not disclose any working examples where an agent, peptide or protein of interest, either conjugated or unconjugated to mutant EtxB/CtxB, is delivered *in vivo* to a target cell in an animal or animal model of disease. The amount of specific guidance required for the claimed method is high because the level of skill in the art and the level of unpredictability in the art are high. For example, Michl et al **(Current Cancer Drug**

Targets, 4:689-702, 2004) teaches the state of art in bacterial toxins as therapeutic agents for cancer. Michl teaches that several hurdles remain in the art of treating cancer with bacterial toxins as immunomodulators and delivery vehicles for antigens/proteins of interest including targeting to cancer cells and maintaining efficacious levels of said antigens/proteins to be delivered to said cancer cells (page 698, col. 1, paragr. 4). Further, the art of record and the teachings of Michl are silent with respect to methods of treating cancer using EtxB/CtxB as a delivery vehicle. The specification fails to provide specific guidance and working examples to address these issues and to enable an artisan to practice the claimed invention commensurate in scope with the instant claims.

The instant claims read on delivery of agents to any target cell. Claim 8 further limits the target cell to an antigen presenting cell (APC). The art of record at the time of the invention is replete with teachings that EtxB/CtxB binds to GM-1 on the surface of cells and that said binding mediates cellular uptake of EtxB/CtxB. For example, Williams et al **(WO00/14114, IDS)** teaches that binding of CtxB/EtxB to GM-1 is a critical event that correlates with CtxB/EtxB entry into a target cell (page 2, lines 1-5; page 4, lines 29-30; page 12, lines 20-25; page 14, lines 7-13). Loregian et al **(Proceedings of the National Academy of Science, USA, 96:5221-5226, 1999)** teaches that CtxB/EtxB binding to GM-1 on the surface on target cells mediates internalization and uptake of CtxB/EtxB (page 5221, col. 1, paragr. 2, lines 5-8). Nashar et al[a] **(Vaccine, 11:235-240, 1993)** teaches that EtxB/CtxB binding to GM-1 mediates the uptake of EtxB/CtxB into eukaryotic cells (page 236, col. 1, paragr. 1). Thus, the art of record at the time of the

invention clearly teaches that CtxB/EtxB can only target cells expressing GM-1. The specification provides no specific guidance or working examples for an artisan to target CtxB/EtxB, or any mutants therof, to any target cell. The specification and the art of record at the time of the invention enable only targeting CtxB/EtxB to cells expressing GM-1.

The instant claims read on CtxB/EtxB mutants containing any mutations within the region spanning amino acid residues E51 to I58 of the β4-α2 loop of EtxB or CtxB. The breadth of the instant claims encompass any point mutation, any mutation that results in truncation of the CtxB/EtxB protein at any one of amino acid residues E51 to I58 or any mutation that deletes one or more amino acids within the region spanning amino acid residues E51 to I58. Dependent claim 10 further limits said mutation to a mutation at amino acid residues 51, 56 and/or 57 of the β4-α2 loop and dependent claim 11 further limits said mutation to wherein the mutant comprises a H57A or H57S mutation. The specification discloses alanine scanning mutagenesis of all residues within the region from E51 to I58 (page 33) but only discloses that EtxB(H57A) covalently conjugated to a 19mer peptide stimulated class I-restricted antigen presentation when incubated with cultured JAWS II cells (page 56). Williams et al **(WO00/14114, IDS)** teaches several CtxB/EtxB mutants including CtxB(E51A), CtxB(V52A), CtxB(P53A), CtxB(G54A), CtxB(S55A), CtxB(Q56A), CtxB(H57A), CtxB(I58A) and EtxB(H57S) (Examples 1 and 2, pages 40-43) but that only molecules with point mutations at three separate sites (positions 51, 56 and 57; i.e. CtxB(E51A), CtxB(Q56A), CtxB(H57A) and EtxB(H57S)) retain GM-1 binding activity (page 12, lines

20-21). It is reiterated, as put forth herein above, that the art of record at the time of the invention clearly teaches that EtxB/CtxB binds to GM-1 on the surface of cells and that said binding mediates cellular uptake of EtxB/CtxB. As such, it is unpredictable whether CtxB/EtxB mutants containing any mutations within the region spanning amino acid residues E51 to I58 of the β4-α2 loop of EtxB or CtxB or whether any mutations at amino acid residues 51, 56 and/or 57 of the β4-α2 loop of EtxB or CtxB would bind GM-1 and be operant in the instantly claimed invention. The specification provides no specific guidance or working examples for an artisan to make and use the claimed invention commensurate with the instant claims.

In summary, an artisan of skill would have required extensive experimentation to practice the claimed invention commensurate in scope with the instant claims. Such experimentation will be undue because of the unpredictability of delivering any peptide or protein of interest to a target cell without conjugating said peptide of protein of interest directly to EtxB/CtxB, because of the unpredictability of targeting EtxB/CtxB to any cell, including cells not expressing GM-1, because of the unpredictability of any mutant EtxB/CtxB binding to GM-1 and because of the unpredictability of treating any disease or disorder, including any viral infection or any cancer, comprising targeting an agent, peptide, protein of interest or antigen to a target cell with a mutant EtxB/CtxB. Neither the specification nor the art of record at the time of the invention provides sufficient guidance to address these issues for an artisan to practice the claimed invention.

Thus, limiting the scope of the claims to:

An *in vitro* method of delivering a peptide to a cell expressing GM-1 ganglioside receptors on the surface of said cell comprising contacting said cell with a mutant B-subunit of *E. coli* heat labile enterotoxin (EtxB) or a mutant B-subunit of *Vibrio cholerae* cholera toxin (CtxB) covalently linked to said peptide, said mutant EtxB or mutant CtxB comprising one of the following point mutations within the region spanning amino acid residues E51 to I58 of the β4-α2 loop of EtxB or CtxB: CtxB(E51A), CtxB(Q56A), CtxB(H57A) and EtxB(H57S), thereby delivering said peptide into said cell; and a kit comprising said mutant EtxB or mutant CtxB covalently linked to said peptide, is proper.

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 1-4, 6-8, 10-18, 23 and 24 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claims 1, 14, 15 and 17 recite methods of "using a mutant of EtxB or CtxB", but, since the claim does not set forth any steps involved in the method/process, it is unclear what method/process applicant is intending to encompass. A claim is indefinite where it merely recites a use without any active, positive steps delimiting how this use is actually practiced. Claims 2-4, 6-8, 10 and 11 depend from claim 1; claim 16 depends from claim 15; and claim 18 depends from claim 17. As such, claims 2-4, 6-8, 10, 11, 16 and 18 are likewise rejected.

Claim 1-4, 6-8, 10-18, 23 and 24 are incomplete. Claims 1, 12, 14, 15 and 17 do not recite any positive steps which clearly relate back to the preamble. Therefore, it is unclear how said steps relate to the preamble recitations of the instant claims and whether the goal of said preamble recitations have been resolved. Claims 2-4, 6-8, 10 and 11 depend from claim 1; claims 13, 23 and 24 depend from claim 12; claim 16 depends from claim 15; and claim 18 depends from claim 17. As such, claims 2-4, 6-8, 10, 11, 13, 16, 18, 23 and 24 are likewise rejected.

Claim Rejections - 35 USC § 102

> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

Claims 1, 2, 10, 11, 17 and 21 are rejected under 35 U.S.C. 102(b) as being anticipated by Williams et al **(WO00/14114, IDS)**.

Williams teaches mutant B-subunits of Etx and Ctx; specifically, CtxB(E51A), CtxB(V52A), CtxB(P53A), CtxB(G54A), CtxB(S55A), CtxB(Q56A), CtxB(H57A), CtxB(I58A) and EtxB(H57S) (Examples 1 and 2, pages 40-43). Said CtxB/EtxB mutants all contain point mutations in the region spanning amino acid residues E51 to I58 of the β4-α2 loop of CtxB/EtxB. Williams teaches that CtxB(E51A) and CtxB(H57A) failed to trigger CD8+ T-cell depletion (Example 1, Results, page 41). Williams teaches that all of said CtxB/EtxB mutants bound to CD8+ T-cells to a similar extent and that both CtxB(H57A) and EtxB(H57S) bound with a slightly higher avidity to GM-1 coated microtitre plates and exhibited a slightly higher K_d for GM-1 as determined by plasmon surface resonance (Example 2, Results, page 43). Williams teaches that mice

immunized intranasally with wild-type EtxB exhibited high titre serum anti-EtxB IgG antibody levels (titre=5757+/-785) while mice immunized intranasally with mutant EtxB(H57S) exhibited significantly lower anti-EtxB IgG antibody levels (titre=1205+/-222) (Example 3, page 43). Williams teaches that both EtxB(H57S) and CtxB(H57A), when coadministered to mice intranasally with ovalbumin, elicited a reduced anti-ovalbumin immunogenic response in said mice when compared to wild-type EtxB and CtxB (Example 4, page 44).

It is noted that Williams is considered to teach "a method of using a mutant of EtxB or CtxB comprising delivering an agent to a target cell" (claim 1), "wherein the agent is selected from the group consisting of a peptide or protein of interest" (claim 2) because Williams teaches a method comprising contacting mouse CD8+ T-cells with the various CtxB/EtxB mutant proteins. Williams teaches that said CtxB/EtxB mutant proteins bound to said mouse CD8+ T-cells and as such, said CtxB/EtxB mutant proteins were delivered to a target cell (i.e. mouse CD8+ T-cells being the target cell). Further, said CtxB/EtxB mutant proteins are considered to be the agent. Thus, Willaims anticipates the instant claims.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

Claims 1-4, 6-8, 10-18, 21, 23 and 24 are rejected under 35 U.S.C. 103(a) as being unpatentable over Williams et al **(WO00/14114, IDS)** in view of Loregian et al **(Proceedings of the National Academy of Science, USA, 96:5221-5226, 1999)**, Marcello **(Proceedings of the National Academy of Science, USA, 91:8994-8998, 1994)**, Nashar et al[a] **(Vaccine, 11:235-240, 1993)** and Nashar et al[b] **(International Immunology, 13:541-551, 2001)**.

The teachings of Williams are put forth herein above under the 35 U.S.C. 102(b) rejection. In addition, Williams teaches that binding of CtxB/EtxB to GM-1 is a critical event that correlates with CtxB/EtxB entry into a target cell (page 2, lines 1-5; page 4, lines 29-30; page 12, lines 20-25; page 14, lines 7-13). Williams teaches that CtxB/EtxB molecules with point mutations within the β4-α2 loop region (i.e. amino acids 45-65) retain GM-1 binding activity but lack other activities, such as immunogenicity and toxicity (page 12, lines 20-25).

Williams does not teach: delivery of a specific agent other than CtxB/EtxB mutant proteins, said agent linked to a membrane translocating or fusigenic peptide, delivery of said agent to a particular compartment of a target cell and delivery of said agent to an antigen presenting cell. Williams also does not teach a method of treating any particular disease or condition by administering to or contacting any particular organism, tissue or cell.

Loregian teaches that a peptide corresponding to the 27 C-terminal amino acids of HSV-1 polymerase (POL), when fused to CtxB/EtxB, mediates delivery of the POL portion of a POL-CtxB/EtxB fusion protein into the nucleus of cells when said POL-

CtxB/EtxB fusion protein is incubated with said cells (page 5223: col. 1 to col. 2, paragr. 2). Loregian teaches that CtxB/EtxB binding to GM-1 on the surface on target cells mediates internalization and uptake of CtxB/EtxB (page 5221, col. 1, paragr. 2, lines 5-8).

Marcello teaches a method of delivery of a peptide into the cytosol of a cell comprising contacting a cell with a CtxB/EtxB protein fused to said peptide (page 8997, col. 2: lines 1-4; paragr. 2, lines 1-6). Marcello teaches that CtxB/EtxB binding to GM-1 on the surface of target cells mediates internalization and uptake of CtxB/EtxB (page 8997, col. 2, lines 1-4). Marcello also teaches that CtxB/EtxB can enter the intracellular vesicular network (page 8998, col. 1, lines 1-4).

Nashar et al[a] teaches the state of the art of EtxB/CtxB as carriers for the oral delivery of heterologous antigens and epitopes. Nashar teaches that EtxB/CtxB are easily manipulated for either genetic or chemical attachment of antigens or epitopes (page 235, col. 2, paragr. 1). Nashar teaches that EtxB/CtxB binding to GM-1 mediates the uptake of EtxB/CtxB into eukaryotic cells (page 236, col. 1, paragr. 1). Nashar teaches that EtxB/CtxB fusion proteins containing proteins of interest to be delivered to a cell can be easily produced in large quantities (page 236, col. 2, paragr. 4 to page 237, col. 1, line 2).

Nashar et al[b] teaches that antigen presenting cells, including B cells and dendritic cells, have GM-1 localized on the cell surface and that said cells can bind EtxB/CtxB (page 544, col. 2, lines 1-13 and see throughout entire document). Nashar teaches that an ovalbumin antigen covalently linked to CtxB/EtxB promotes uptake and

immunological response to ovalbumin in said cells. Nashar teaches that CtxB/EtxB binding to GM-1 on the surface on target cells mediates internalization and uptake of CtxB/EtxB (page 549, col. 1, line 1 to paragr. 3).

Loregian, Marcello, Nashar et al[a] and Nashar et al[b] do not teach mutants of EtxB/CtxB containing mutations in the region spanning amino acid residues E51 to I58.

Thus, Williams teaches EtxB/CtxB mutants that possess GM-1 binding activity and that exhibit reduced immunogenicity and reduced toxicity compared to wild type EtxB/CtxB. Marcello and Loregian teach that EtxB/CtxB can be fused to peptides or proteins of interest as a means to deliver said peptides or protein of interest into target cells expressing GM-1. Marcello teaches that EtxB/CtxB proteins facilitate delivery specifically to the cytosolic compartments of target cells while Loregian teaches that EtxB/CtxB can be further modified by fusing the C-terminal region of HSV-1 polymerase to EtxB/CtxB resulting in delivery of said peptides or proteins of interest specifically to the nuclear compartment of target cells. Williams, Loregian, Marcello, Nashar et al[a] and Nashar et al[b] all teach that is was well known at the time of the invention that binding of ExtB/CtxB to GM-1 receptors on the surface of target cells was predictive of entry of said ExtB/CtxB into said target cell. As such, it would have been obvious to an artisan of ordinary skill at the time of the invention to combine the teachings of Williams, Loregian, Marcello, Nashar et al[a] and Nashar et al[b] with a reasonable expectation of success. That is, an artisan of ordinary skill would have had a reasonable expectation of success to combine the teachings of Williams with those of Loregian, Marcello, Nashar et al[a] and Nashar et al[b] to arrive at a method of delivering a protein of interest to an antigen

presenting cell comprising providing a fusion protein consisting of a mutant EtxB/CtxB (i.e. a mutant EtxB/CtxB as taught by Williams) covalently linked to said protein of interest, wherein said mutant EtxB/CtxB is further modified by covalently linking POL as taught by Loregian (i.e. for nuclear delivery of said protein of interest) and contacting any cell with GM-1 present on the cell surface, thereby delivering said protein of interest into said cell.

An artisan of ordinary skill would have been motivated to combine the teachings of Williams, Loregian, Marcello, Nashar et al[a] and Nashar et al[b] as such because Williams teaches that CtxB/EtxB mutants containing point mutations in the region spanning amino acid residues E51 to I58 of the β4-α2 loop of CtxB/EtxB exhibit reduced toxicity. Loregian provides motivation to utilize a CtxB/EtxB-based peptide/protein delivery system as a means to target particular cellular compartments. Loregian recites (page 5226, col. 1, paragr. 4):

> The ability of EtxB to enter the endocytotic pathway, in which a proportion of the material traffics to the Golgi, suggests that EtxB can find widespread application as a delivery system. EtxB-based chimeras might be engineered by introduction of specific signals into attached peptides to either enhance or prevent cleavage, potentiate or inhibit insertion across endocytic membranes, and ensure targeting to other intracellular compartments, such as Golgi apparatus, endoplasmic reticulum, or mitochondria. Our demonstration that the Pol peptide, when delivered by EtxB, retains its pharmacological characteristics heralds the prospect that B subunit-peptide conjugates could be used as a screen for bioactive peptides that inhibit intracellular enzymes and macromolecular complexes relevant to the pathogenesis of microbial infections and malignant diseases. One could also envisage, as an *in vivo* application of this system, the intracellular delivery of immunodominant epitopes as a conceptually new type of vaccines that exploit the intrinsic, immune-adjuvant activity of EtxB and its mucosal tropism (35, 36).

Nashar et al[b] teaches that CtxB/EtxB, coupled to a protein of interest, can be delivered to antigen presenting cells and that an immunological response to said protein of interest can be elicited. Thus, an artisan of ordinary skill would have been motivated to combine the teachings of Williams, Loregian, Marcello, Nashar et al[a] and Nashar et al[b] because the mutant CtxB/EtxB taught by Williams exhibited reduced toxicity which would be advantageous when delivering a protein of interest to any cell, including antigen presenting cells and when delivering a protein of interest to any compartment of said cell.

Please note that intended use limitations bear little weight on the determination of patentability. In the instant case, for claims 1, 12, 14, 15, 17 and 21, the limitations "a method of using a mutant EtxB or CtxB", "a method of preparing a medicament", "a method of using a mutant of EtxB or CtxB for separate, simultaneous or combined use

to treat a disease or a condition in a subject in need of same", "a method of treating a disease or condition in a subject in need of same", "a method of delivering an agent using a mutant to a target cell" and "a kit for delivering an agent to a target cell", respectively, does not carry patentable weight in the determination of anticipation for the claimed products or processes. This is because a recitation of the intended use of the claimed invention must result in a structural difference between the claimed invention and the prior art. In a claim drawn to a process, the intended use must result in a manipulative difference as compared to the prior art. See MPEP § 2111.02, *In re Casey*, 152 USPQ 235 (CCPA 1967) and *In re Otto*, 136 USPQ 458, 459 (CCPA 1963). In the instant case, since the method steps recited in the claim(s) and the method steps taught by the combined teachings of Williams, Loregian, Marcello, Nashar et al[a] and Nashar et al[b] are the same, practice of the process would inherently result in the same outcome.

Thus, the claimed invention as a whole was *prima facie* obvious.

Conclusions

No claims are allowed.

If Applicants should amend the claims, a complete and responsive reply will clearly identify where support can be found in the disclosure for each amendment. Applicants should point to the page and line numbers of the application corresponding to each amendment and provide any statements that might help to identify support for the claimed invention (e.g. if the amendment is not supported *in ipsis verbis*, clarification on the record may be helpful). Should Applicants present new claims, Applicants should clearly identify where support can be found in the disclosure.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Paul Dowell whose telephone number is 571-272-5540. The examiner can normally be reached on M-F, 8-4:30.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ram R. Shukla, can be reached on 571-272-0735. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

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